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repeats of retroviruses, promoters of long-terminal repeats of transposable elements, the Simian Virus 40 early promoter, the cytomegalovirus immediate early promoter, the adenovirus major late promoter,] and a Hepatitis B viral promoter, wherein said gene is operably linked to said promoter.

#### REMARKS

##### The Invention

The invention provides a method for expressing an exogenous gene in a mammalian cell by introducing into a mammal containing the cell a baculovirus containing the exogenous gene. The invention also provides a nucleic acid that includes a recombinant baculovirus genome, an exogenous mammalian gene, and an exogenous mammalian promoter or Hepatitis B viral promoter.

##### Pending Claims

Claims 1 and 21-26 are pending in the application, with claims 1 and 26 having been amended. Claim 19 has been cancelled, and the limitations of claim 19 have been incorporated into claim 1. No new matter has been added.

##### Double Patenting

Applicant acknowledges the provisional double patenting rejection of claim 1 pending the issuance of U.S. Serial No. 08/311,157 as a patent. This provisional rejection has been

obviated by incorporation of the limitations of claim 19 into claim 1 by the above amendment.

35 U.S.C. § 112, ¶ 1

Claims 1, 19, and 21-25 were rejected under 35 U.S.C. § 112, first paragraph, for lack of enablement. Applicant respectfully traverses this rejection.

The rejection is predicated on the ground that the specification does not provide an enabling disclosure for expression of exogenous genes *in vivo* such that the infected cell will be alive and will provide treatment of a gene deficiency disease. In reaching this conclusion, the Examiner stated:

The specification has shown some *in vitro* expression of a marker gene  $\beta$ -galactosidase in HepG2 cells, and very poor expression in 3T3 mouse fibroblasts and in human hepatocytes SKHep1, which clearly suggests that not all cells are capable of expressing exogenous genes at a high level. The specification has shown somewhat better expression in only HepG2 cells because these cells contain asialoglycoprotein receptor.... [Office Action, pg. 4; emphasis added.]

As an initial matter, Applicant respectfully points out that neither the claims of the application nor the nature of the invention requires that the exogenous genes are expressed at a "high level." Thus, it is improper to read this limitation into the claims.

In reference to the substance of the rejection, Applicant respectfully points out that it is incorrect to conclude that "not all cells are capable of expressing exogenous

genes at a high level." Applicant has clearly asserted that "[n]early all mammalian cells are potential targets of the AcMNPV and other baculoviruses" (page 10, lines 18-20), and this assertion is supported by the data set forth in the specification and the accompanying Declaration of Dr. Frederick M. Boyce ("the Declaration"), the inventor of the claimed subject matter.

The Declaration demonstrates that the methods described in the specification can be used to express exogenous genes in a wide variety of mammalian cell types. The experiments summarized in the Declaration demonstrate exogenous gene expression in the following cell types: HepG2, 293, PC12 (treated with nerve growth factor), HeLa, CHO/dhfr<sup>-</sup>, C<sub>2</sub>C<sub>12</sub> myoblasts, C<sub>2</sub>C<sub>12</sub> myotubes, primary human myoblasts, Sk-Hep-1, NIH3T3, NIH3T3 (expressing an asialoglycoprotein receptor), Hep3B, FT02B, Hepa 1-6 cells, neurons, glia, skin, spleen, kidney, stomach, skeletal muscle, uterus, and pancreas.

These examples also demonstrate that expression of an exogenous gene in Sk-Hep-1 cells, for example, can be achieved merely by increasing the dosage (i.e., multiplicity of infection) of the virus administered to the cell. The experiments described at page 7 of the Declaration show that an exogenous gene can be expressed in Sk-Hep-1 cells simply by administering baculovirus at a multiplicity of infection (moi) of 125. This moi is well within the range of moi disclosed in the specification as being useful for achieving exogenous gene expression in nearly all mammalian cells (an moi of 0.1 - 1,000 is recommended at page 16, lines 12-13).

It is improper to reject the claims on the belief that "better expression [was shown] in only HepG2 cells because these cells contain asialoglycoprotein receptor" (Office Action, pg. 4; emphasis added). Applicant respectfully reminds the Examiner that "it is not a requirement of patentability that an inventor correctly set forth, or even know, how or why the invention works." *Newman v. Quigg*, 877 F.2d 1575 (Fed. Cir. 1989). Furthermore, the specification cannot be fairly read to assert that a high level of expression was seen in HepG2 cells only because those cells have an asialoglycoprotein receptor. Instead, the specification clearly asserts that exogenous gene expression can be achieved *in vitro* or *in vivo* in a wide variety of mammalian cell types, including cells that lack the asialoglycoprotein receptor.

While experiments described in the specification and in the accompanying Declaration demonstrated expression of the enzyme  $\beta$ -galactosidase, nothing in the Office Action lends support to the notion that genes other than  $\beta$ -galactosidase (e.g., therapeutic genes) could not be expressed using the claimed baculovirus expression system.

The accompanying Declaration, for example, shows that the claimed baculovirus expression system can be used to express the therapeutic human liver enzyme ornithine transcarbamylase in human liver cells. Given these data and Applicant's data showing *in vivo* expression of exogenous genes, one skilled in the art can have a reasonable expectation of success in expressing a therapeutic gene *in vivo* as claimed.

The Office Action states that "the specification has not taught what is the level of expression of the desired therapeutic gene in vivo, viability of the infected cells in vivo, and correlation between the expression of the desired gene and the disease treatment" (Office Action, pg. 5). Applicant disagrees. Achieving a suitable level of gene expression can be readily accomplished simply by adjusting the dosage of virus as desired (see, e.g., page 17, lines 14-24 of the specification). Indeed, any level of gene expression can be expected to ameliorate a gene deficiency disorder; a complete cure is not necessary to satisfy the enablement requirement.

In sum, no undue experimentation would be required of a person of ordinary skill in the art who wished to practice the claimed invention. In view of (a) the extensive evidence that the claimed invention can be used with a wide variety of mammalian cell types *in vivo*, (b) the evidence that therapeutic genes can be expressed in mammalian cells, and (c) the evidence that such use involves a straightforward application of the guidance provided in the specification, as asserted in the specification, this rejection for lack of enablement should be withdrawn.

35 U.S.C. § 103

Claims 1, 19, and 26 were rejected as being unpatentable for obviousness over Miller U.S. Patent No. 5,004,687 in view of Fraser (Curr. Topics in Microbiol.

Immunol. 158:131-172, 1992) and Srivastava U.S. Patent

No. 5,252,479. The rejection is predicated on the belief that:

It would have been obvious ... to use the baculovirus vectors of Miller for expressing mammalian genes as taught by Fraser with the expectation to get increased expression of mammalian genes and without any pathogenic effects to the host cells as taught by Miller.... There would be a reasonable expectation of success given the data of Miller and Fraser. [Office Action, pg. 7.]

This rejection is overcome by the above amendment to claim 26 and the remarks set forth below.

With respect to the claims as amended, this rejection is flawed. The prior art as a whole simply would not have motivated one skilled in the art to produce the claimed invention because the prior art failed to provide a reasonable expectation of success in using the claimed invention to express exogenous genes in mammalian cells. Indeed, the prior art teaches away from the invention.

Briefly, prior to this inventor's work, baculovirus-based vectors were not thought to be useful for directing gene expression in mammalian cells. Baculovirus-based vectors containing exogenous genes operably linked to mammalian promoters, as claimed, would have been thought to be useless. Therefore, prior to this inventor's work, one would not have been motivated to combine a baculovirus genome with a mammalian promoter, as it would not have been expected to direct exogenous gene expression in either mammalian or insect cells. Because

"both the suggestion and the expectation of success must be founded in the prior art, not in the applicant's disclosure," this rejection for obviousness should be withdrawn. *In re Dow Chemical Co.*, 837 F.2d 469 (Fed. Cir. 1988).

A review of the prior art shows that baculoviruses would not have been expected to direct exogenous gene expression in mammalian cells. While the Miller patent may be read as asserting that baculoviruses containing a Rous Sarcoma Virus long terminal repeat (RSV-LTR) can express genes in insect and mammalian cells, the Miller patent must be read in light of all of the prior art.

In determining whether such a suggestion can fairly be gleaned from the prior art, the full field of the invention must be considered; for the person of ordinary skill is charged with knowledge of the entire body of technological literature, including that which might lead away from the claimed invention. [Dow at 473; emphasis added.]

When the entire body of technological literature is considered, it is clear that the prior art teaches away from the claimed nucleic acids.

In a paper published after the 1985 filing date of the Miller patent, Miller retracted the assertions relied upon by the Examiner. In her 1987 paper entitled "Baculovirus Interaction with Nontarget Organisms: a Virus-Borne Reporter Gene is Not Expressed in Two Mammalian Cell Lines," Miller unambiguously retracted her previous conclusions (Carbonell and Miller, 1987, Appl. and Environ. Microbiol. 53:1412-1417 (cited as reference AK in the Information Disclosure Statement submitted on

February 26, 1997)). Based upon experiments employing the same mouse cell line described in the Miller reference and an additional human cell line, Miller concluded in 1987 that:

the reporter gene CAT under the control of a mammalian-active promoter, when carried as part of the DNA genome of AcMNPV, is not expressed in two inoculated mammalian cell lines. [Carbonell and Miller at 1416; emphasis added.]

As Miller noted, her 1987 conclusions are consistent with previous reports that baculoviruses do not direct gene expression in mammalian cells. In 1987, Miller reported:

Results of this study corroborate those of previous reports that affirm the safety of baculoviruses with respect to vertebrates, especially mammals, and show that although AcMNPV may gain entry into the cell, the viral DNA either does not reach the nucleus or is not available to cell transcriptional factors.... When [the reporter gene] gains access to the cells as part of the AcMNPV genome, [the reporter gene] is not expressed. All accumulated evidence suggests that insect baculoviruses pose no threat to nontarget organisms. [Carbonell and Miller at 1416; emphasis added.]

Thus, Miller ultimately withdrew the assertions relied upon by the Examiner.

The claims of the Miller patent are limited to the use of insect cells, rather than mammalian cells. Thus, although a patent is presumed to provide a valid enabling disclosure for its claims, no such presumption exists for mere assertions embedded in the specification and to which no claims are directed.



The Fraser reference cited in the Office Action does not make up for the deficiencies of Miller. If anything, the Fraser reference suggests that the host range of baculoviruses is limited to insects:

The insect-pathogenic nuclear polyhedrosis viruses ... replicate within the nuclei of susceptible insect cells. [Fraser at 132-133.]

Fraser certainly fails to teach or suggest that baculoviruses can be used to express an exogenous gene in a mammalian cell.

As mentioned above, the cited references must be considered in light of the full field of technical knowledge. Dow at 473. As a review of the prior art shows, those of ordinary skill in the art would not have expected baculoviruses to direct gene expression in mammalian cells. Rather, the art as a whole teaches away from the invention. For example, Blissard and Wenz taught:

Baculoviruses infect invertebrates exclusively. [Blissard and Wenz, 1992, J. Virol. 66:6829-6835 (cited as reference AG in the Information Disclosure Statement submitted on February 26, 1997).]

Likewise, after contacting mammalian cells with baculoviruses, Gröner et al. reported:

[T]he available data strongly suggest that AcNPV [i.e., baculovirus] does not replicate in vertebrate cell cultures and that this virus is specific for several lepidopterous insect species. [Gröner et al., 1984, Interviol. 21:203-209 at 208 (cited as

reference BF in the Information Disclosure Statement submitted on February 26, 1997).]

Hartig et al. also concluded that baculoviruses are unable to infect vertebrate cells:

AcNPV-exposed [i.e., baculovirus-exposed] cells did not exhibit cytopathic changes indicative of infection, nor was infectious virus produced from exposed cells. [Hartig et al., 1991, J. Virol. Meth. 31:335-344 at 339 (cited as reference BH in the Information Disclosure Statement submitted on February 26, 1997).]

In a review article on baculoviruses, Blissard and Rohrmann also suggest that the host range of baculoviruses is limited to insects and a few crustacea:

The Baculoviridae are a family of occluded DNA viruses pathogenic predominantly for holometabolous insects.... Baculoviruses are a diverse group of large viruses ... which are pathogenic for invertebrates. While the vast majority of baculoviruses infect insects ... several have also been found in crustacea. [Blissard and Rohrmann, 1990, Ann. Rev. Entomol. 35:127-155 at 127 (submitted as reference AF in the Information Disclosure Statement submitted on February 26, 1997).]

Dimmock and Primrose also teach that baculoviruses infect only insect cells:

Of the eight groups of insect viruses described so far only the baculoviruses appear to be restricted to insects and because of their specificity they have been considered as biological control agents. [Dimmock and Primrose, *Introduction to Modern Virology*, Blackwell Scientific Publications,

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Oxford, England, 1987, pg. 304 (copy enclosed as Exhibit A).]

The 1996-97 catalogue of CLONTECH corporation, which markets baculovirus vectors, also teaches away from the invention. According to CLONTECH Corporation, the supposed inability of baculoviruses to infect mammalian cells is an advantage of the virus:

Safe - baculoviruses only replicate in insect cells; they do not infect humans, animals, or plants [CLONTECH catalogue, 1996-1997, pg. 118; emphasis added (copy enclosed as Exhibit B).]

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As the above excerpts illustrate, the art actually taught away from the present invention at the time the present application was filed in 1994 (and to some extent even continues to do so today). It is improper to read the Miller, Fraser, and Srivastava patents in isolation. As the *Dow* court mandated, the full field of the invention must be considered. Included within this field is Miller's 1987 paper that retracts the conclusions upon which the Examiner relied. This 1987 paper plus the numerous other publications described above establish that in 1994 those of ordinary skill in the art would certainly not have expected that baculoviruses could direct exogenous gene expression in mammalian cells.

Because baculoviruses were not thought to direct exogenous gene expression in mammalian cells, a person of ordinary skill in the art would not have been motivated to

produce the claimed nucleic acids. As amended, the claims are limited to nucleic acids that include a mammalian promoter or a Hepatitis B viral promoter, and methods for expressing exogenous genes in mammalian cells. The recited promoters are active in mammalian cells, but they are thought not to be active, or at least not to work efficiently, in insect cells. Given that, prior to Applicant's discovery, baculoviruses were not thought to direct gene expression in mammalian cells, a person of ordinary skill in the art would not have been motivated to combine a genome of a virus that was thought to be insect-specific with a promoter that is thought to function efficiently in mammalian cells, but not in insect cells. None of the cited references lends support to the contrary.

Thus, the prior art failed to provide (i) the requisite motivation to combine the elements of the claims, and (ii) a reasonable expectation of success in practicing the invention. In view of these remarks and the above amendment, this rejection for obviousness should be withdrawn.

Claims 21-25 were rejected as being unpatentable for obviousness over Miller U.S. Patent No. 5,004,687, Fraser (Curr. Topic. Microbiol. Immunol. 158:131-172, 1992), Grompe et al. (Adv. Exp. Med. and Biol. 3098:51-56, 1991), and Wilson et al. (J. Biol. Chem. 267:963-967, 1992). This rejection has been obviated by the above amendment and remarks set forth herein.

The Miller and Fraser references are relied upon by the Examiner as disclosing the use of baculovirus vectors for


expressing mammalian genes in mammalian cells. The Grompe and Wilson references are relied upon as disclosing gene therapy methods for treating gene deficiency disorders in hepatocytes. This rejection is flawed for the reasons given above, and it is not rescued by the Grompe and Wilson references. Even when Grompe and Wilson are considered, the prior art as a whole taught away from the invention, and a person of ordinary skill in the art would not have had a reasonable expectation of success in expressing exogenous genes in mammalian cells. Thus, the skilled artisan would not have been motivated to produce the claimed nucleic acids - regardless of which gene is expressed. In view of these remarks and the above amendment, this rejection for obviousness should be withdrawn.

CONCLUSION

Applicant submits that all of the pending claims are now in condition for allowance, which action is requested. Filed herewith is a Petition for Automatic Extension, along with the required fee. Please charge any additional fees, or apply any credits, to Deposit Account No. 06-1050.

Respectfully submitted,

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